

CLAIMS

1. An isolated mesenchymal stromal stem cell (MSSC) that has been differentiated *in vitro* towards, or to, an intervertebral disc (IVD) cell phenotype for use as a medicament.
2. An isolated mesenchymal stromal stem cell (MSSC) characterised in that it is:
 - a) differentiated *in vitro* towards, or to, a intervertebral disc (IVD) cell phenotype; and
 - b) genetically transformed with an exogenous gene which codes for a protein that reduces degeneration of an intervertebral disc.
3. The isolated mesenchymal stromal stem cell according to claim 1 or 2 wherein the cell produces an extracellular matrix.
4. The isolated mesenchymal stromal stem cell according to claim 3 wherein the extracellular matrix is identifiable as an IVD extracellular matrix and is distinguishable from an extracellular matrix produced by a chondrocyte.
5. The isolated mesenchymal stromal stem cell according to claim 4 wherein the IVD matrix is characterised by at least one:
 - (a) aggrecan gene expression is greater than collagen type II gene expression;
 - (b) the proteoglycan versican is expressed; or
 - (c) the GAG: hydroxproline ratio (i.e proteoglycan : collagen ratio) is greater than 10:1.
6. The isolated mesenchymal stromal stem cell according to any preceding claim that is derived from blood, bone marrow, or adipose tissue.
7. The isolated mesenchymal stromal stem cell according to claim 6 that is derived from bone marrow in the sternum, femur or iliac crest.
8. The isolated mesenchymal stromal stem cell according to any preceding claim wherein the MSSCs are differentiated using any one of the steps:

- (a) growth in a IVD cell induction medium containing TGF β , CDMP1 or CDMP2;
- (b) encapsulation of the MSSC;
- (c) application of Load to the MSSCs;
- (d) Co-culture of MSSCs with Nucleus Pulposus cells/IVD cells;
- (e) Culture of the MSSCs in conditioned media in which IVD cells have previously been grown;
- (f) Culture in low oxygen tensions; or
- (g) Genetically transformed using a gene regulator of IVD cell differentiation

9. The isolated mesenchymal stromal stem cell according to claim 8 wherein differentiation is effected by using any combination of steps (a), (b), (c), (d), (e) (f) and (g) .

10. The isolated mesenchymal stromal stem cell according to claim 9 wherein the MSSCs are differentiated by encapsulating MSSCs in a gel; and growing the encapsulated cells in a medium for up to 5 weeks during which time a cyclical load equivalent to that experienced in vivo is exerted using hydraulic or other methodology

11. The isolated mesenchymal stromal stem cell according to claim 10 wherein the media is an induction medium according to claim 8(a).

12. The isolated mesenchymal stromal stem cell according to claim 10 wherein the media is a conditioned medium according to claim 8(e).

13. The isolated mesenchymal stromal stem cell according to claim 10 wherein the MSSCs are co-cultured with cells according to claim 8(d).

14. The isolated mesenchymal stromal stem cell according to any one of claims 11 - 13 wherein the oxygen pressure is reduced to less than 5% of the atmosphere in which the cells are cultured.

15. The isolated mesenchymal stromal stem cell according to any one of claims 2- 14 wherein the exogenous gene may be selected from the group of genes encoding proteins involved in the regulation of inflammation and the group comprises genes encoding cytokines; inhibitors of cytokines; and inhibitors of degradative enzymes

16. The isolated mesenchymal stromal stem cell according to any one of claims 2- 15 wherein exogenous gene encodes Interleukin 1 Receptor Antagonist (IL-1RA).

17. A use of a cell according to any one of claims 1 –16 in the manufacture of a medicament for the treatment of spinal conditions characterized by degeneration of the intervertebral disc.

18. The use according to claim 17 wherein the spinal condition is Low Back Pain, degeneration of the intervertebral disc, age-related changes of the intervertebral disc or spondylolysis.

19. The use of a cell according to claims 17 or 18 wherein the cells are for direct injection into an IVD exhibiting DIVD.

20. The use of a cell according to claims 17 or 18 wherein the cells are for seeding onto or into biomaterial scaffolds or gels.

21. A method of treating spinal conditions characterized by degeneration of the intervertebral disc comprising administering to a diseased intervertebral disc of a subject in need of such treatment an isolated MSSC that has been differentiated *in vitro* towards, or to, an IVD cell phenotype.

22. A method of treating spinal conditions characterized by degeneration of the intervertebral disc comprising administering to a diseased intervertebral disc of a subject in need of such treatment an isolated MSSC, wherein said MSSC has been has been:

- (a) differentiated *in vitro* towards, or to, a IVD cell phenotype; and
- (b) genetically transformed with an exogenous gene which codes for a protein that reduces degeneration of an intervertebral disc.

23. A method for causing mesenchymal stromal stem cells to differentiate towards IVD cells comprising exposing cultured mesenchymal stromal stem cells to increasing pressures of up to 30 psi (2.1MPa).

24. A method for causing mesenchymal stromal stem cells to differentiate towards IVD cells comprising co-culturing NP cells and mesenchymal stromal stem cells (MSSCs) together.

25. A method for causing mesenchymal stromal stem cells to differentiate towards IVD cells comprising culturing mesenchymal stromal stem cells in media that has previously been exposed to NP cells.
26. A method for causing mesenchymal stromal stem cells to differentiate towards IVD cells comprising culturing mesenchymal stromal stem cells in an atmosphere in which oxygen pressure is reduced to less than 5%.
27. A method for causing mesenchymal stromal stem cells to differentiate towards IVD cells comprising encapsulating MSSCs in a gel and growing the encapsulated cells in a medium for up to 5 weeks during which time a cyclical load equivalent to that experienced in vivo is exerted using hydraulic or other methodology
28. The method according to claim 27 wherein the media is an induction medium as defined in claim 8(a).
29. The method according to claim 27 wherein the media is a conditioned medium as defined in claim 8(e).
30. The method according to claim 27 wherein the MSSCs are co-cultured with cells according to claim 8(d).
31. The method according to any one of claims 27 –30 wherein the oxygen pressure is reduced to less than 5% of the atmosphere in which the cells are cultured.